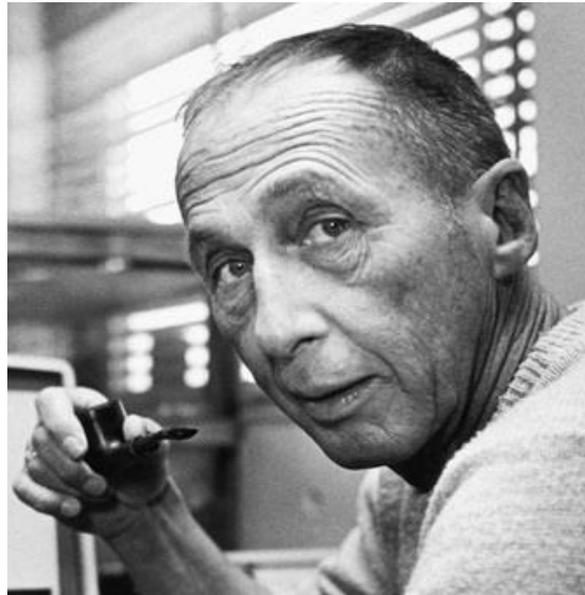
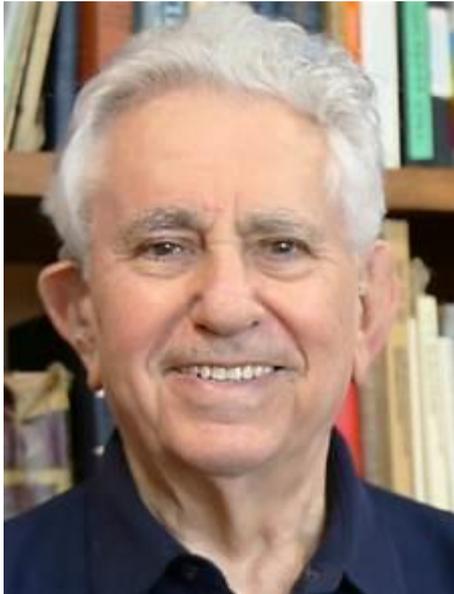
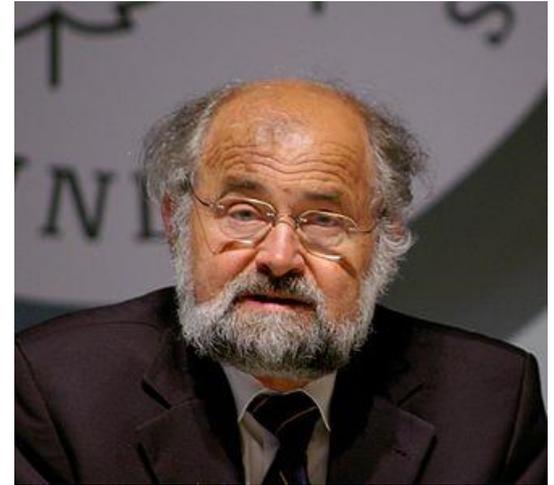
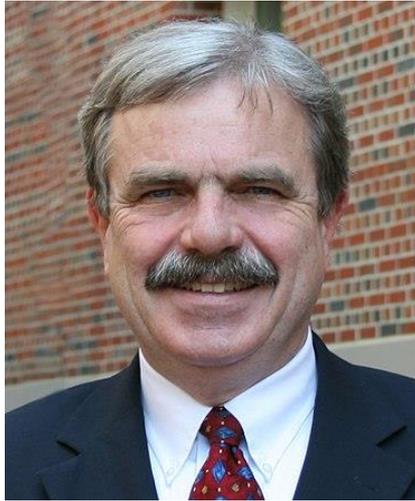


Next Generation Applications of Bioelectrochemistry



Shelley D. Minteer
Department of Chemistry
University of Utah

Bioelectrochemistry Today is Standing on the Shoulders of Giants

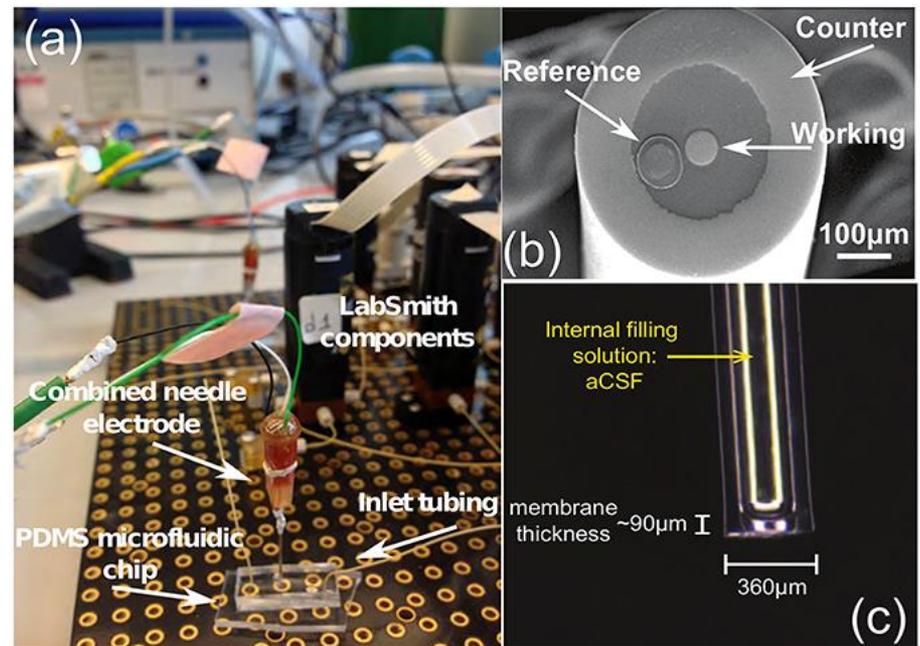


Bioelectrochemistry

- Neuroelectrochemistry
- Biosensors
- Wastewater treatment
- Biofuel cells/Energy Harvesting
- Biobatteries
- Bioelectrosynthesis

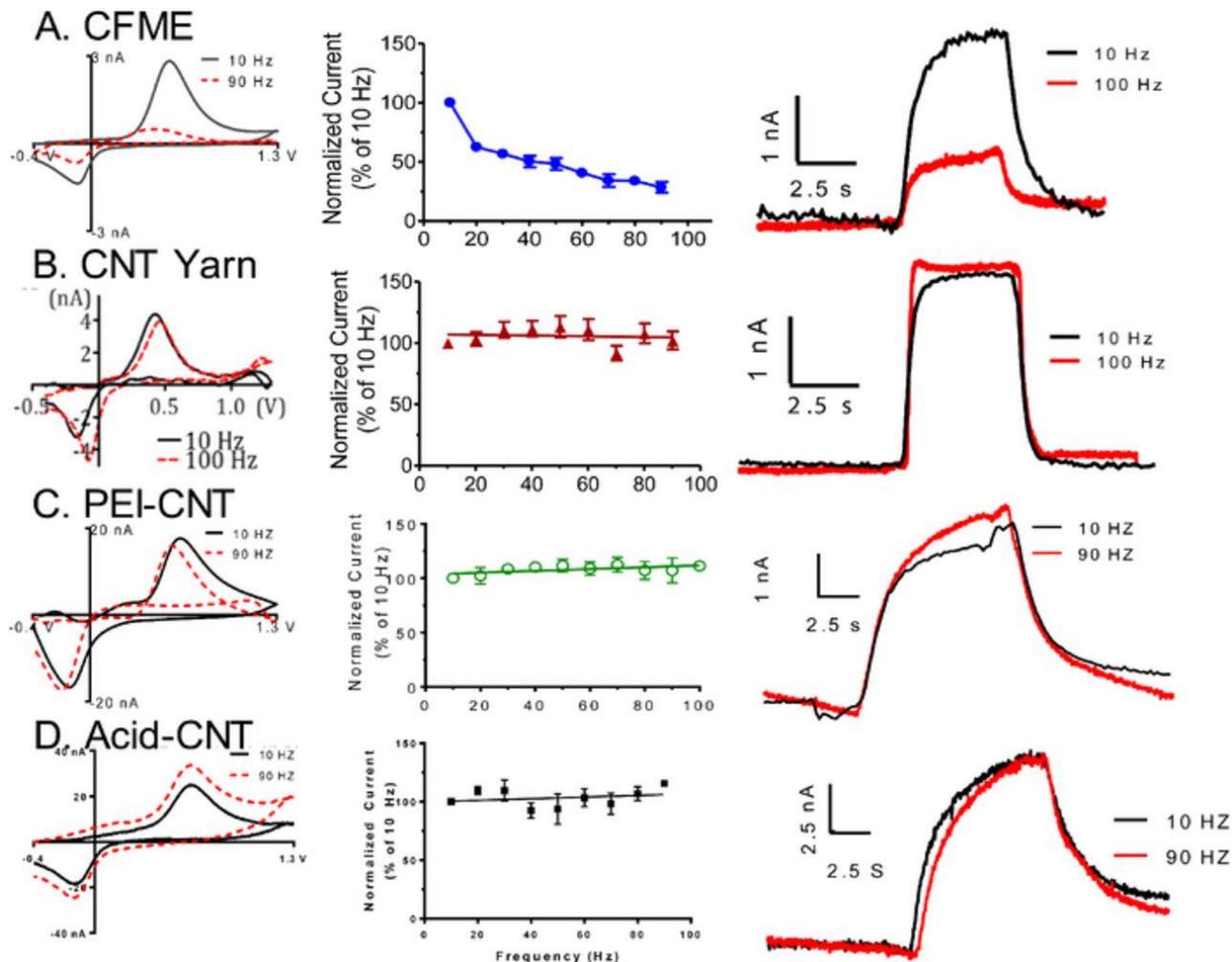
Neuroelectrochemistry

- Type of analysis
 - In-vivo
 - Single neuron
- Analytes
 - Dopamine
 - Potassium
 - Glutamate
 - Nitric oxide
 - Etc.



Papadimitriou KI, Wang C, Rogers ML, Gowers SAN, Leong CL, Boutelle MG and Drakakis EM (2016) High-Performance Bioinstrumentation for Real-Time Neuroelectrochemical Traumatic Brain Injury Monitoring. *Front. Hum. Neurosci.* 10:212. doi: 10.3389/fnhum.2016.00212

Fast-scan voltammetry of neurotransmitter release

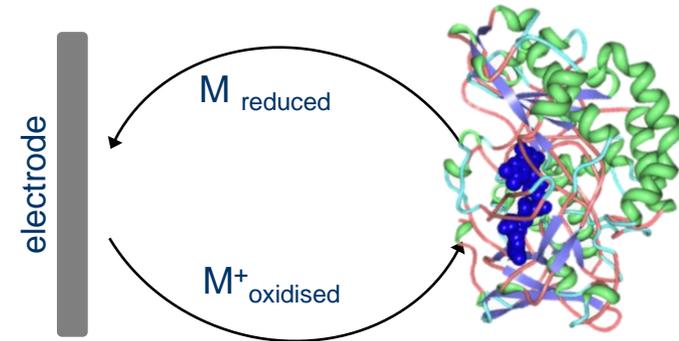


Alexander G. Zestos, and B. Jill Venton J. Electrochem. Soc. 2018;165:G3071-G3073

Bioelectrocatalysis of Enzymes and Microbes

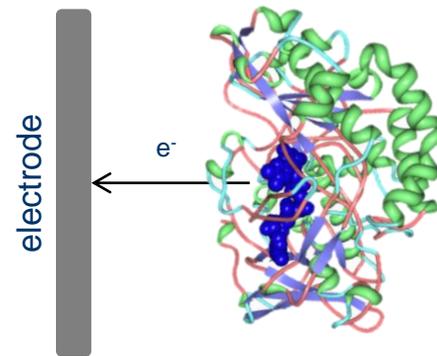
- Mediated Electron Transfer (MET)

- Diffusive, redox-active species shuttles electrons between enzyme and electrode
- Fe-containing, Os-containing, ABTS, quinone-derivatives



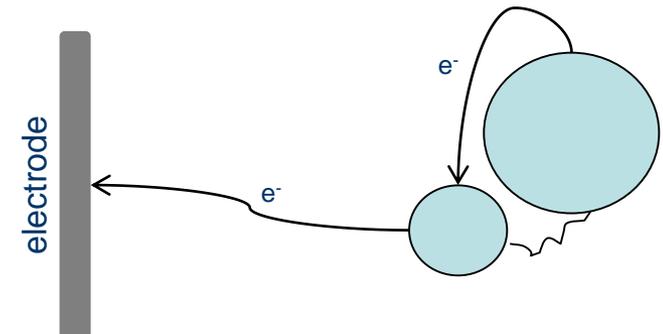
- Direct Electron Transfer (DET)

- Electron tunnels directly between enzyme and electrode (**restrictions apply**)



- Inter-domain Electron Transfer (IET)

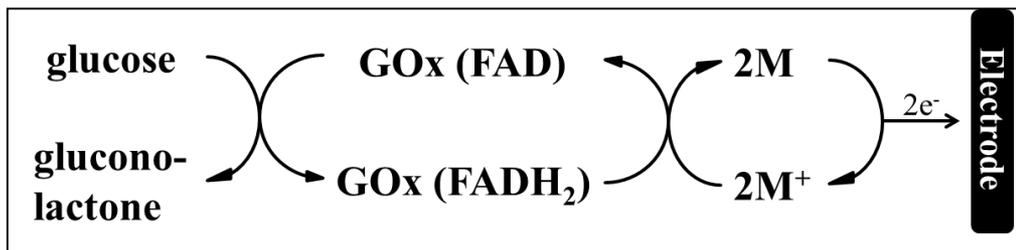
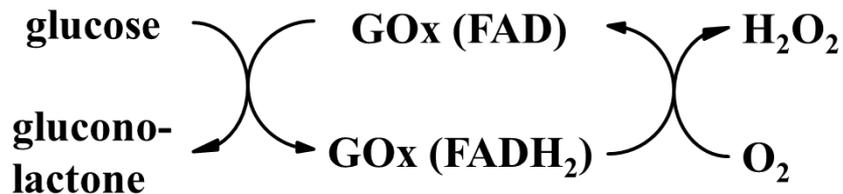
- Main domain (DET impossible) shuttles electron using tethered, sub-domain (DET) possible (usually heme moiety)



Bioelectrocatalysis of Glucose Oxidase

From Biosensors to Biofuel Cells to Bioelectrosynthesis

- 1st gen. = measure O_2 depletion by GOx
 - Significant variation in $[O_2 \text{ dissolved}]$
- 2nd gen. = detect H_2O_2 at an electrode (electrooxidation of H_2O_2)
 - Interferents can be oxidized under same conditions (false positive) = ascorbic acid, acetaminophen
- **3rd gen. = transport electrons from $FADH_2$ to an electrode (mediation)**
 - **Mediators need to be immobilized to prevent leaching, especially for implantable devices**
- 4th gen. = *direct electron transfer from $FAD/FADH_2$ to electrode*
 - Cofactor buried too deeply for direct electron transfer
 - Commonly mistaken for oxygen reduction/depletion



Biosensors

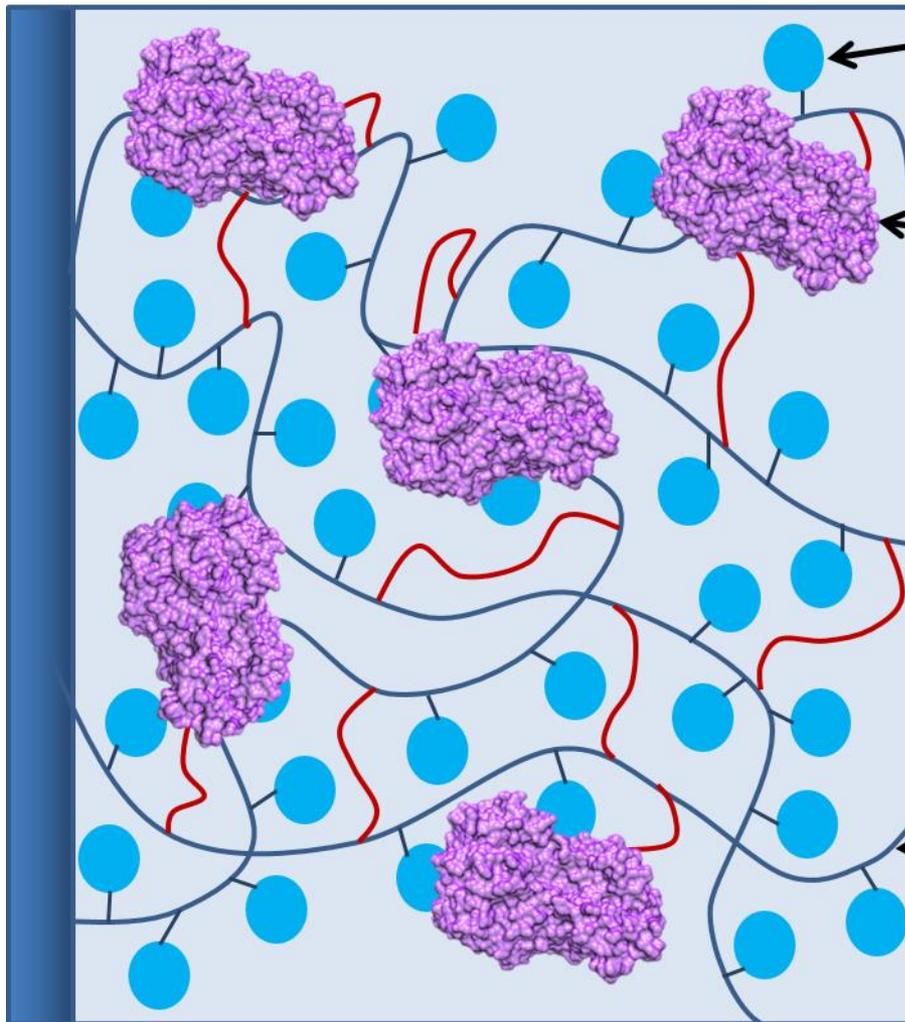
Analytes

- Glucose
- Lactate
- Peroxide
- Pesticides
- Nerve agents
- DNA
- Bacteria, fungi, virus

Blood glucose monitoring
-\$11B market in the US in 2024



Many design variables....



Mediator:

- Redox potential
- Self-exchange rate affects the current

Enzyme:

- Cofactor redox potential affects mediator choice
- 3D structure
- Specific activity

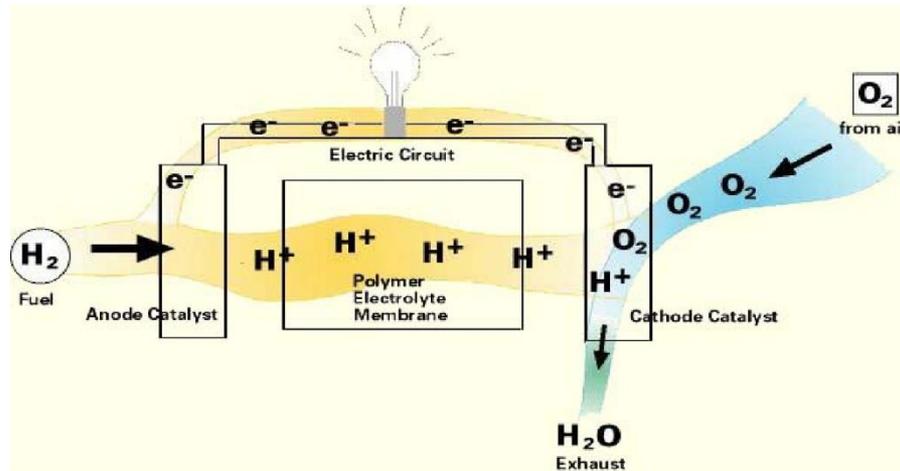
Crosslinker:

- Diffusion through the network
- Separation between mediator and active site

Polymer:

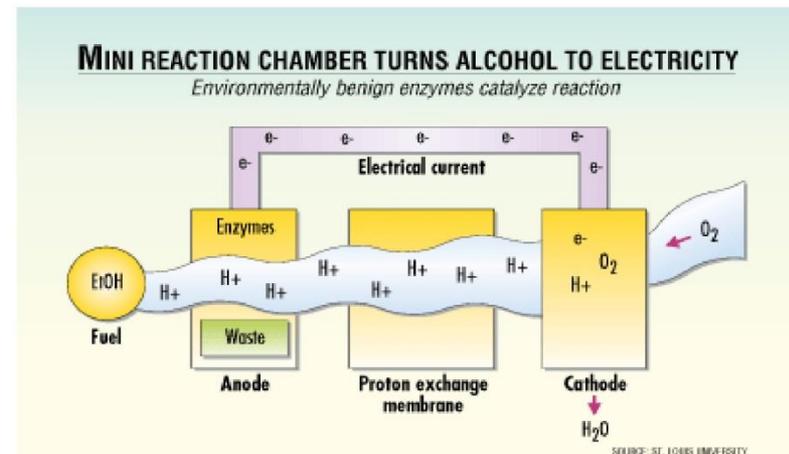
- Diffusion through the network

Hydrogen/O₂ Fuel Cell vs Biofuel Cell



Traditional Hydrogen Fuel Cell

Ethanol Biofuel Cell



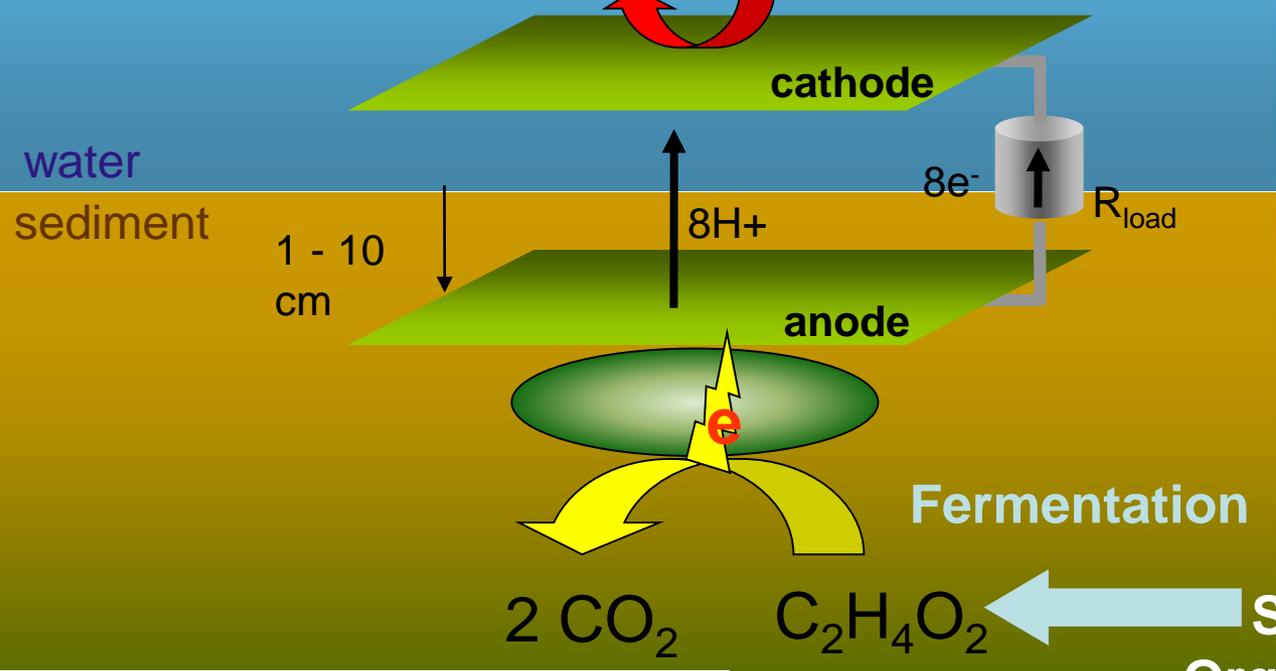


Microbial Fuel Cells

***Geobacteraceae* Can Use Electrodes as an Electron Acceptor
Harvesting Power From Aquatic Sediments**

Cathode Reaction:
 $2O_2 + 8H^+ + 8e^- \rightarrow 4H_2O$

Sediment Battery

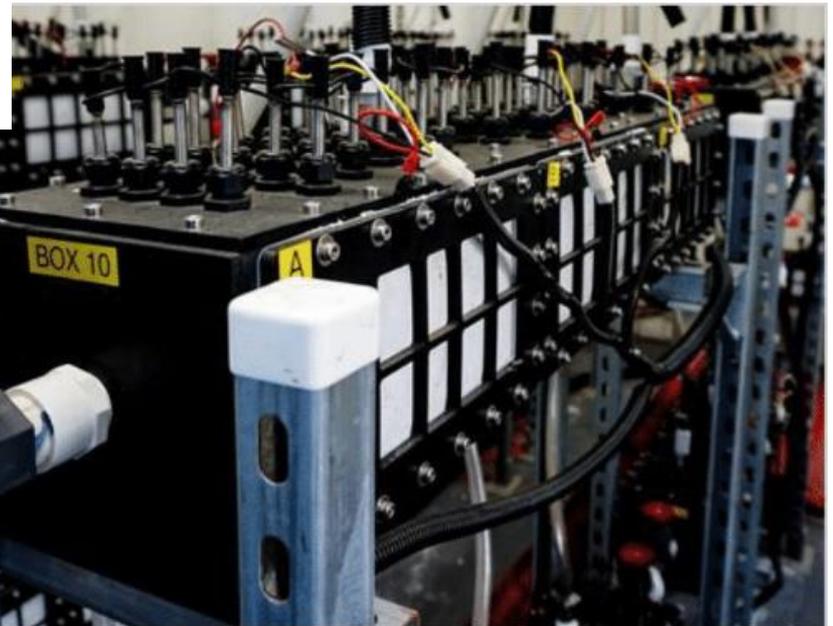
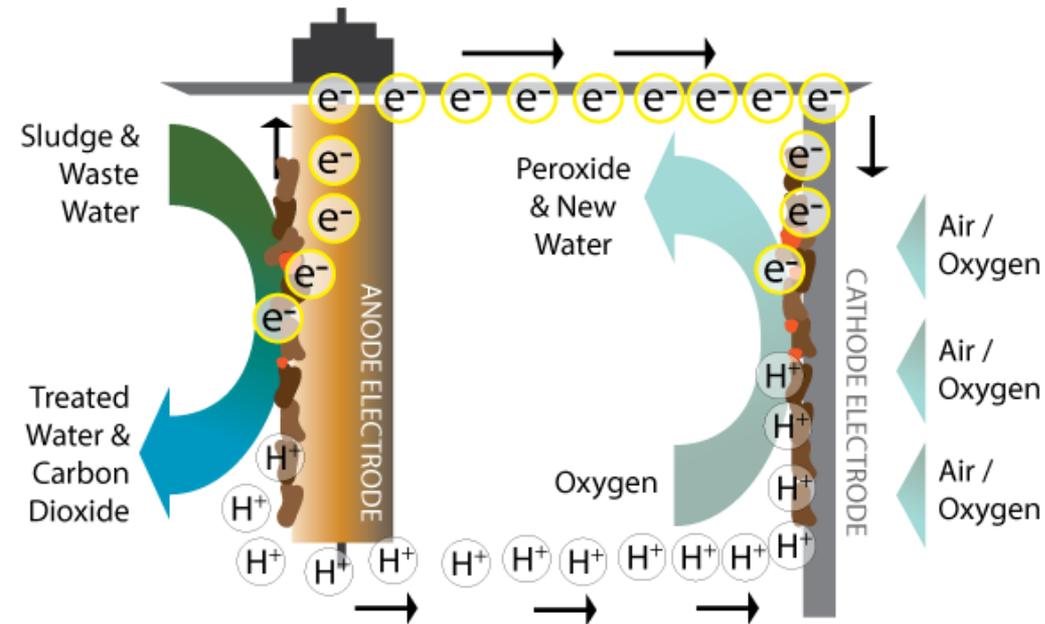


Anode Reaction:
 $C_2H_4O_2 + 2H_2O \rightarrow 2CO_2 + 8H^+ + 8e^-$

**Sediment
Organic Matter**



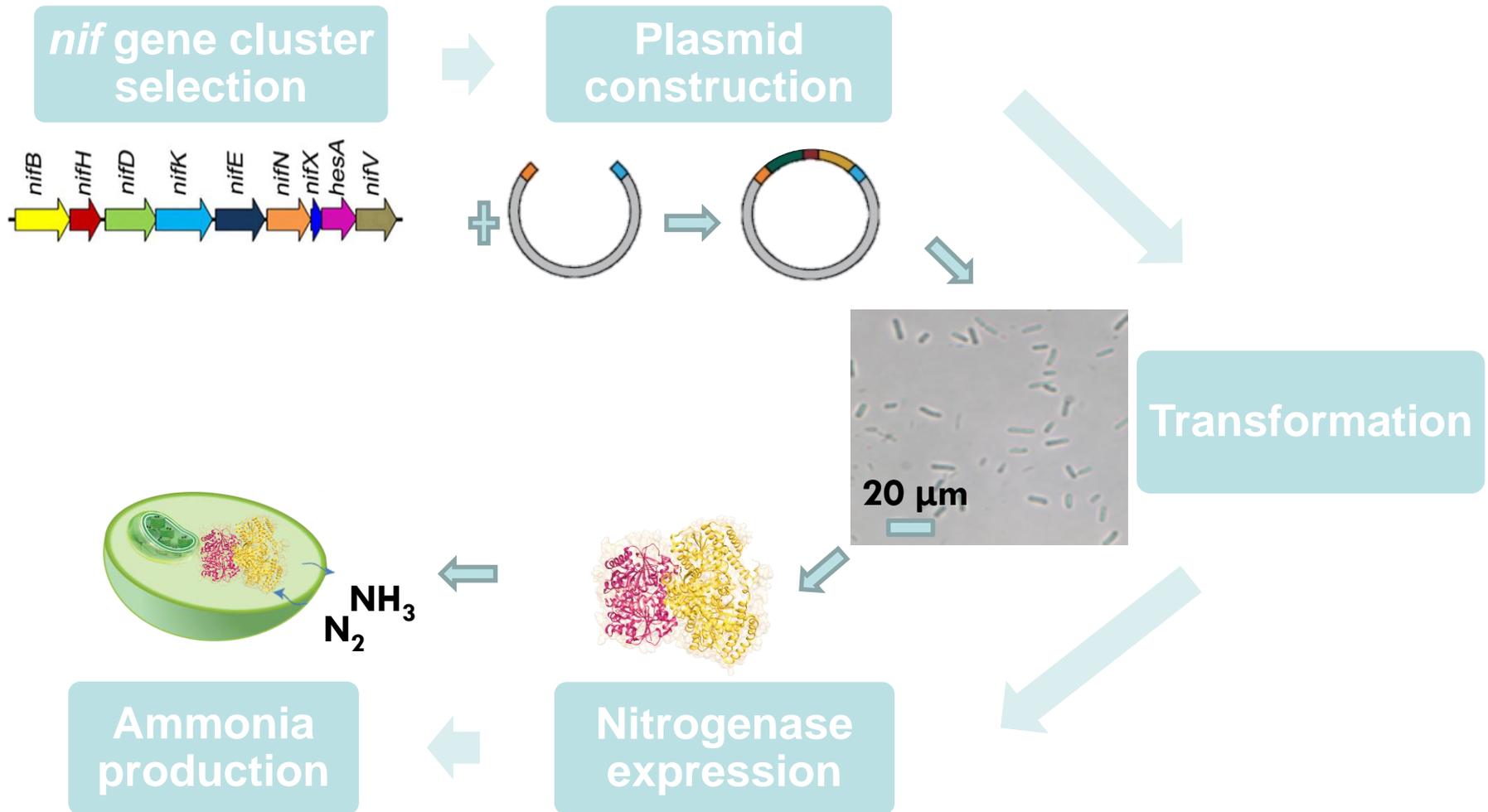
AQUACYCL



Types of Microbial Fuel Cells

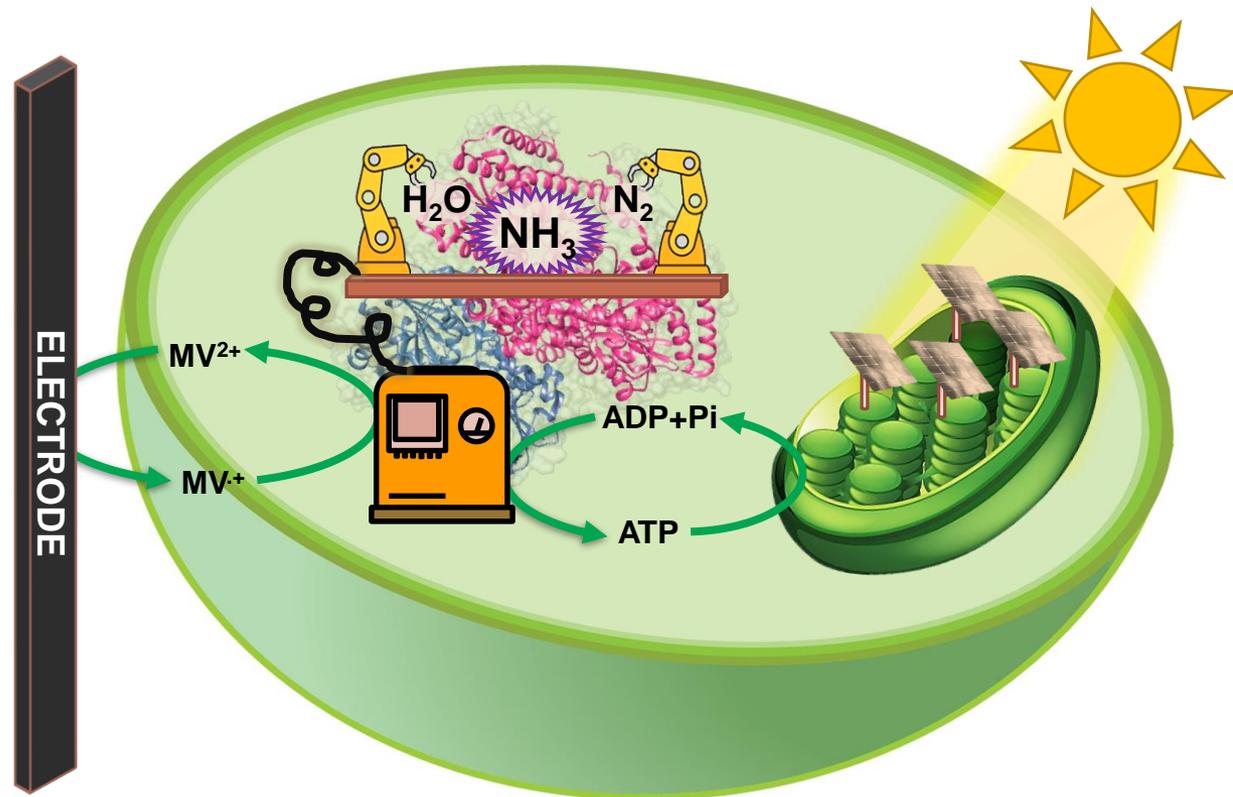
- Electricity production from the oxidation of organic material (in waste) – energy efficient wastewater treatment
- Hydrogen or methane can be produced at biocathodes with externally enhanced potential (BES- Bioelectrochemical systems)
- Synthetic biology will allows us to move beyond this to other chemical products

Engineered *S. elongatus* pcc 7942 for Nitrogen Fixation

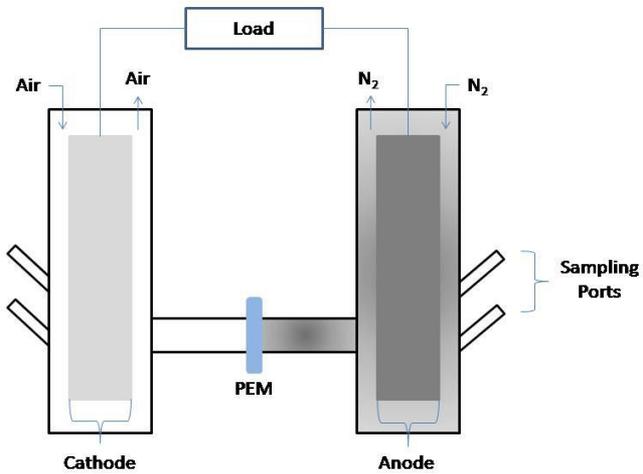


Unicellular factory for ammonium production

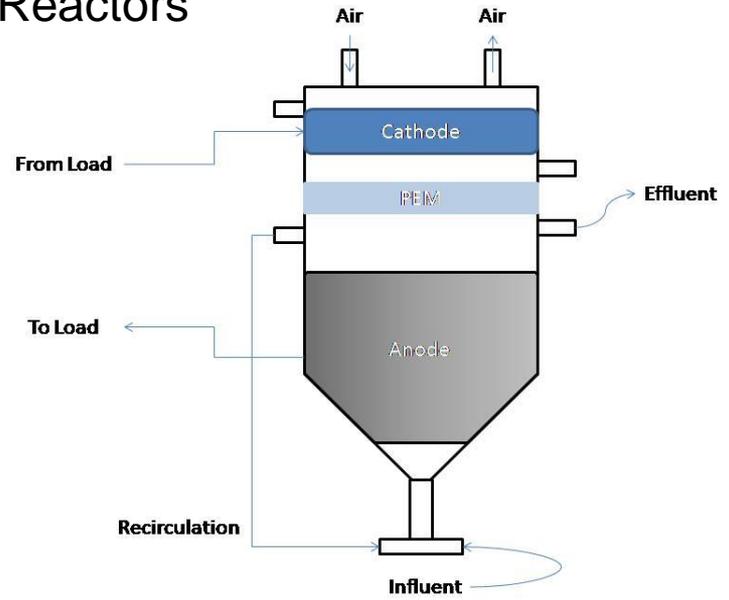
- ❖ Whole cell biocatalysis
- ❖ High enzyme stability
- ❖ Endogenous ATP



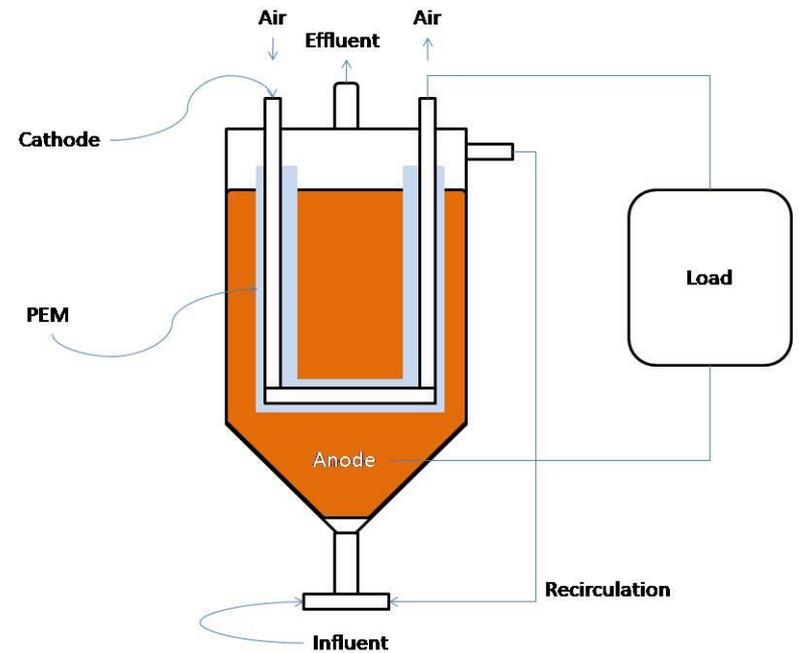
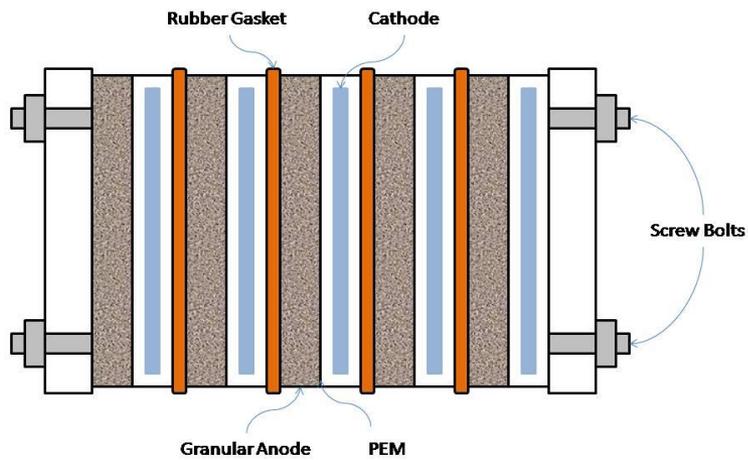
H-cell or U-cell



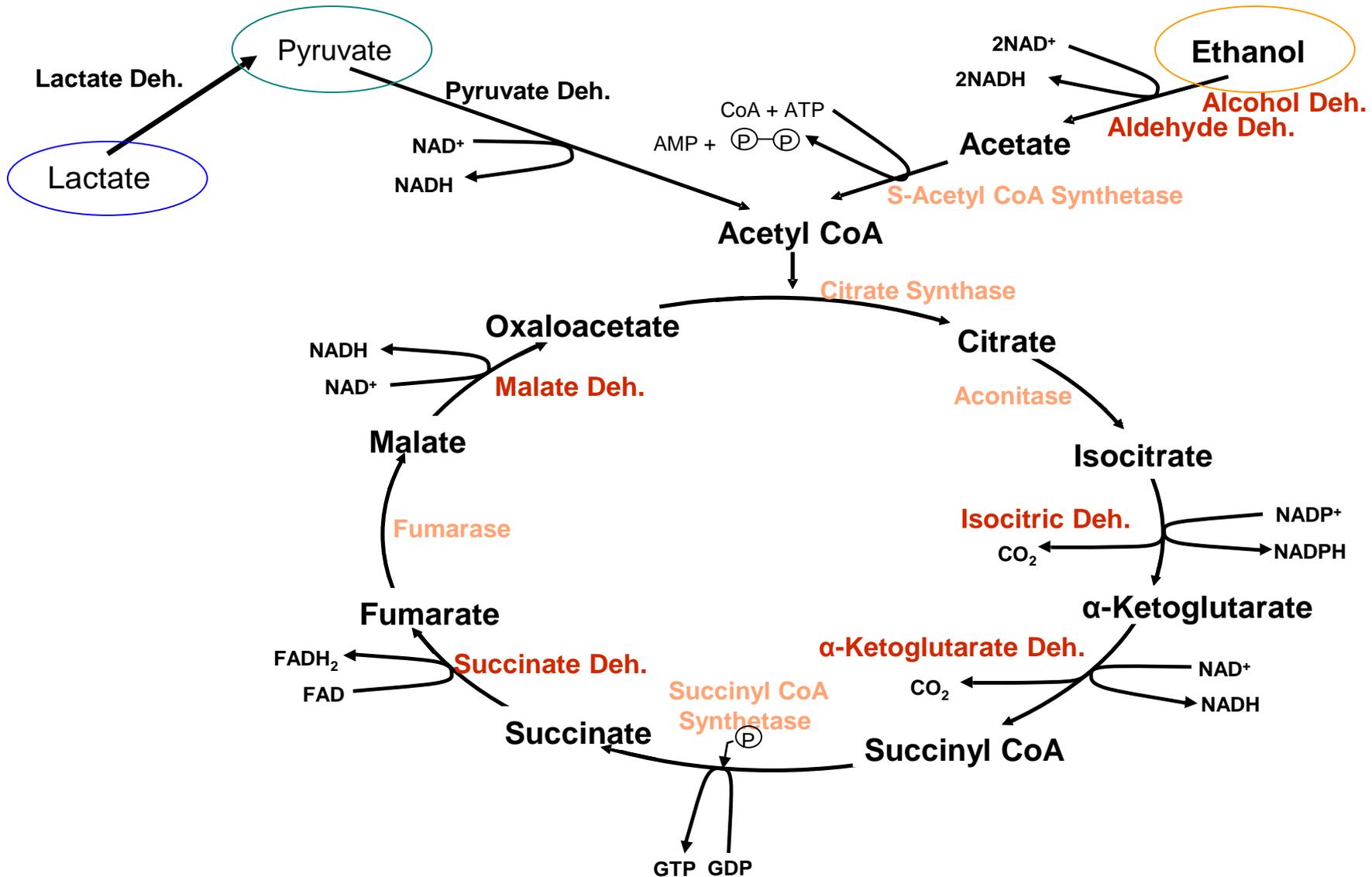
Reactors



Stacks

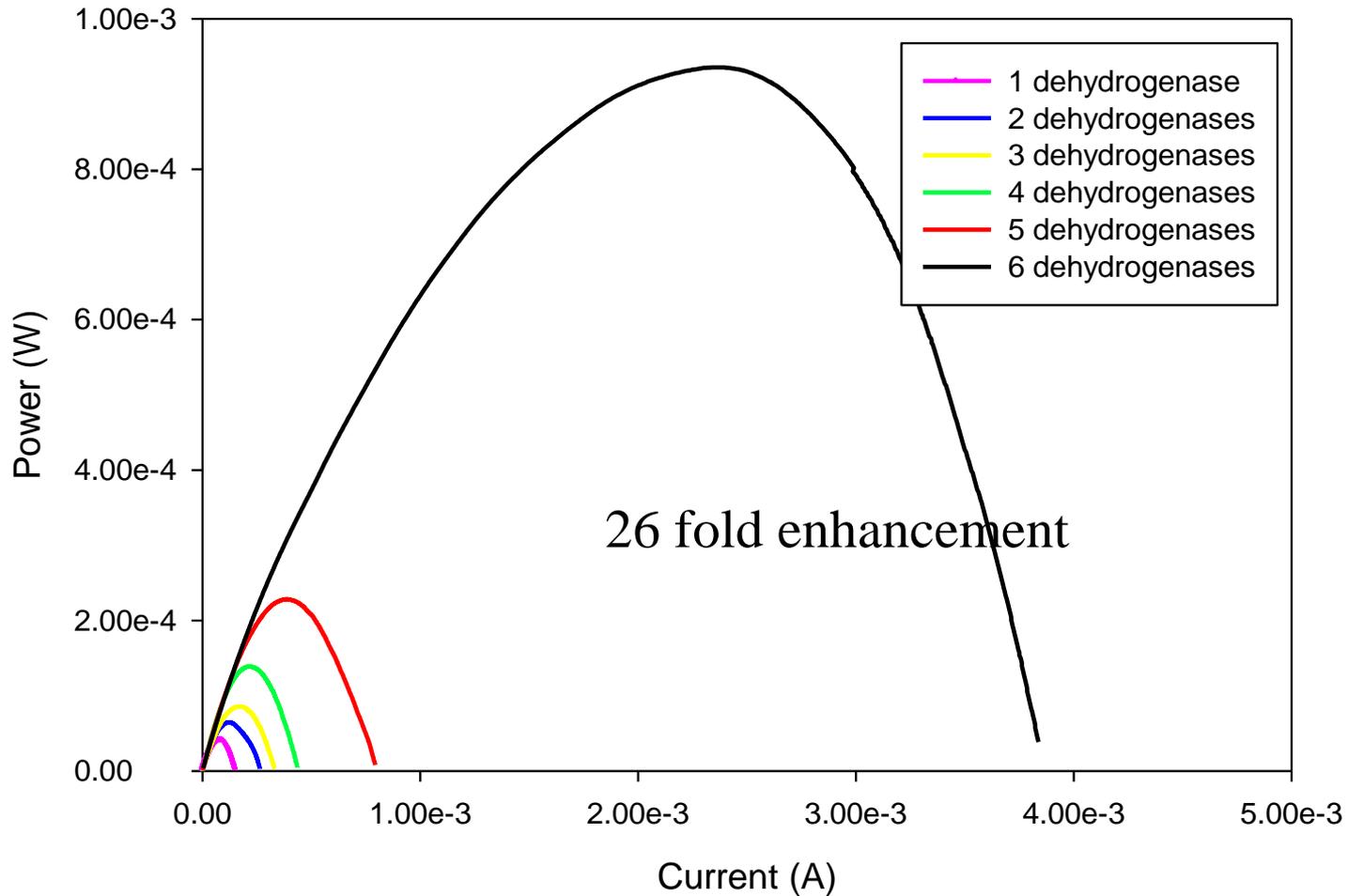


Citric Acid Cycle Biomimic



Citric acid cycle biomimic

Fuel: Lactate

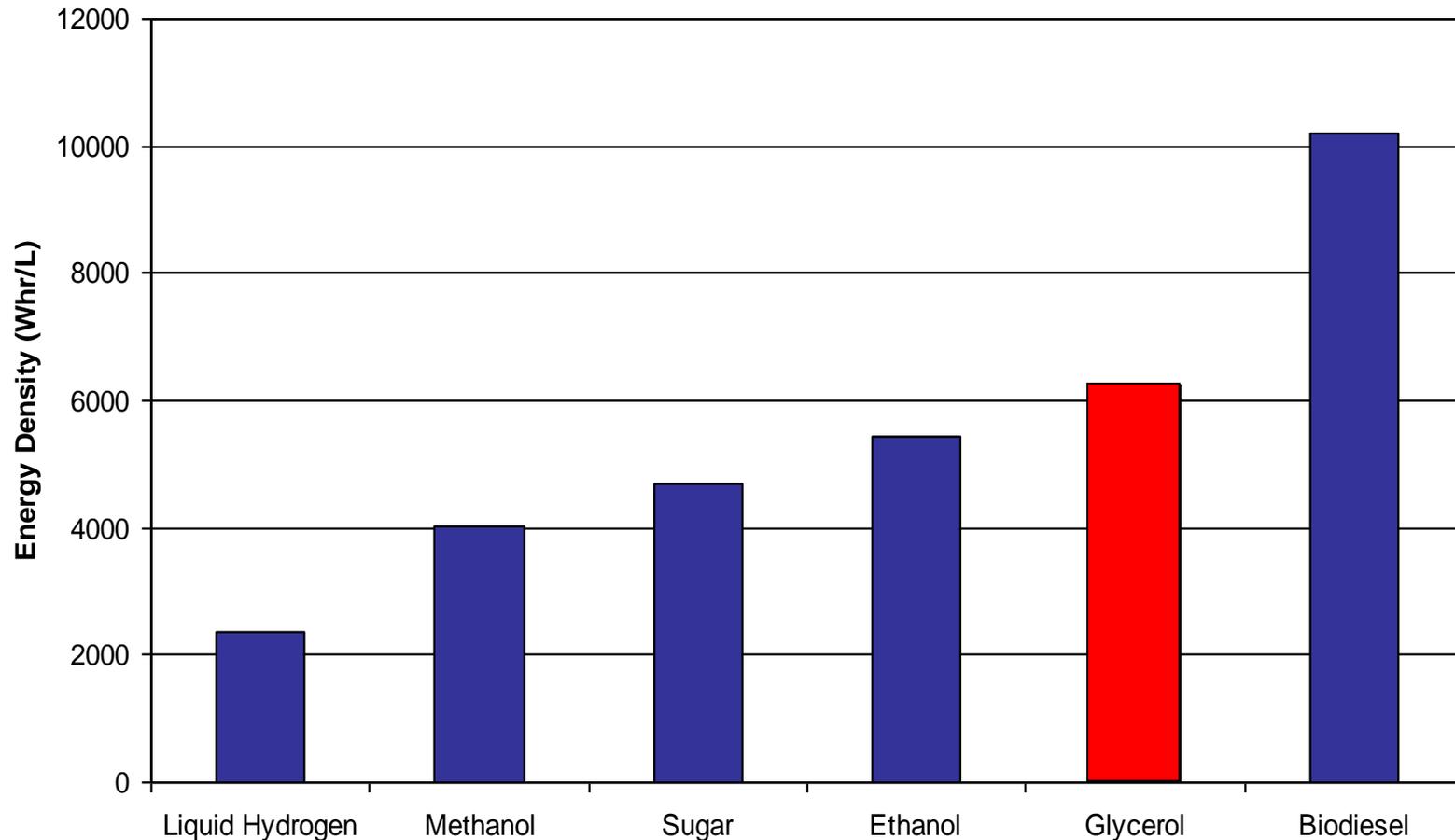


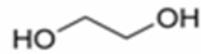


Non-natural Enzyme Pathways: Glycerol as a Fuel of Choice

- ❑ **Byproduct of biodiesel - 10% wt.**
- ❑ **High energy density**
- ❑ **Low volatility**

- ❖ **Inexpensive**
- ❖ **Widely available**
- ❖ **Nontoxic**

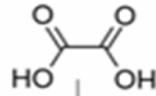




Alcohol Dehydrogenase



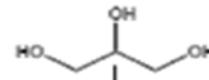
Aldehyde Dehydrogenase



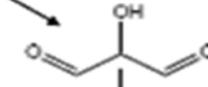
Oxalate Oxidase

2 CO₂

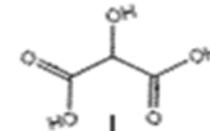
Ethylene Glycol



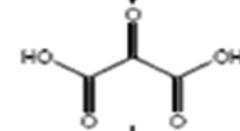
Alcohol Dehydrogenase



Aldehyde Dehydrogenase

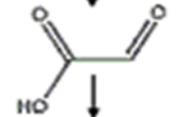


Alcohol Dehydrogenase

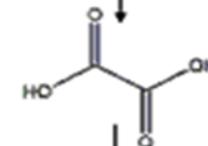


Oxalate Oxidase

CO₂



Aldehyde Dehydrogenase



Oxalate Oxidase

2 CO₂

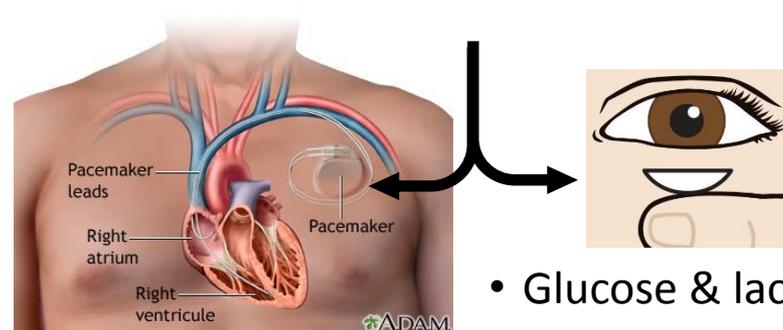
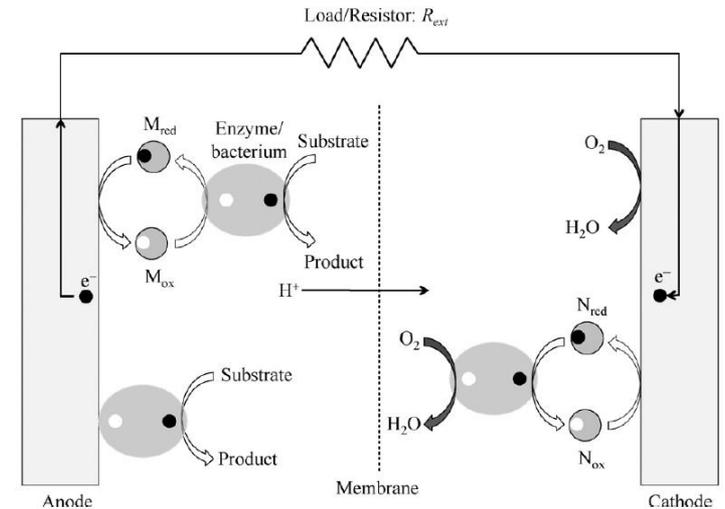
Glycerol

Glycerol Oxidation

Energy Harvesting for Wearables

Our 'Lab on the Skin' Journey

Joseph Wang
UCSD



- Glucose & lactate sensor
- Intraocular pressure (IOP) monitor
- Presbyopia correction
- Electronic display

<http://joewang.ucsd.edu/>

Osman, M.H., et al, *Biosens. Bioelectron.* **26** (2011) 3087–3102

<http://www.fda.gov/ForConsumers/ConsumerUpdates/ucm318020.htm>

<http://www.nlm.nih.gov/medlineplus/ency/imagepages/19566.htm>

Contact lens energy harvesting challenges

Adhesion

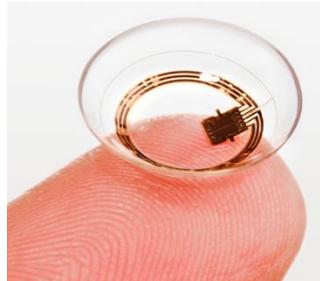


Pt film on plastic



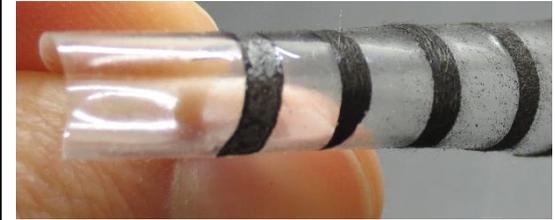
Buckypaper on elastomer

Reactant Accessibility



Electrodes inside elastomer

Flexibility



AvCarb carbon paper on PDMS

Conductivity

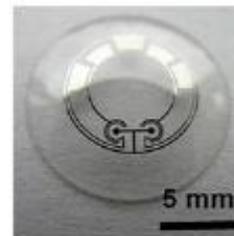


Carbon ink on PDMS

Biocompatibility

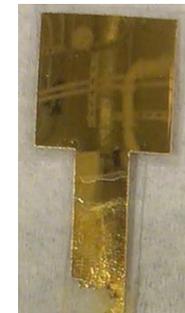


Carbon ink + oil on PDMS



Lactate sensor on plastic lens

Surface Area

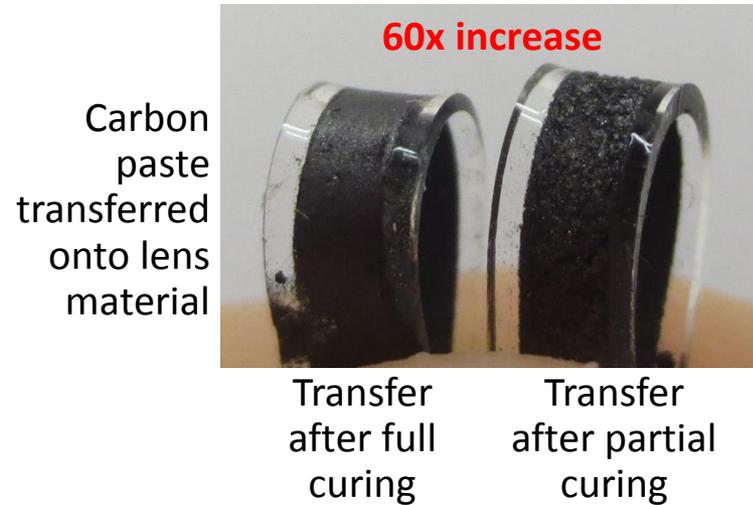


Gold film on plastic

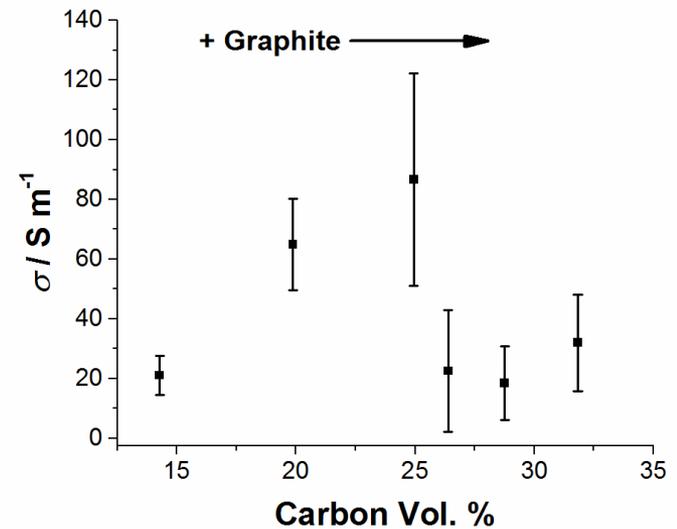
Electrode improvements

	Flexible	Adheres to lens	Biocompatible	High Surface Area	High Conductivity
Buckypaper	✓			✓	✓
AvCarb carbon paper				✓	✓
Metal film	✓		✓		✓
Graphite / PDMS	✓	✓	✓		
Graphite / PMMA			✓		
Graphite / polystyrene			✓		
Graphite / chitosan			✓		
Ercon carbon ink			✓		

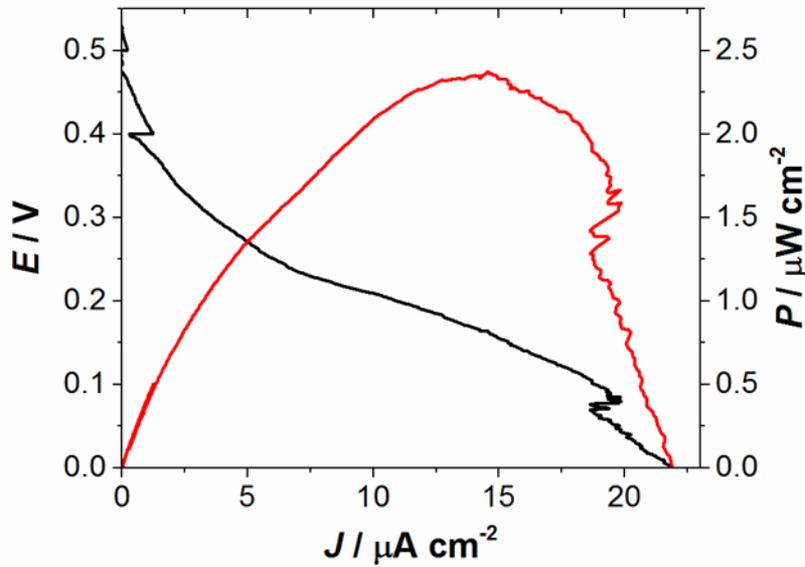
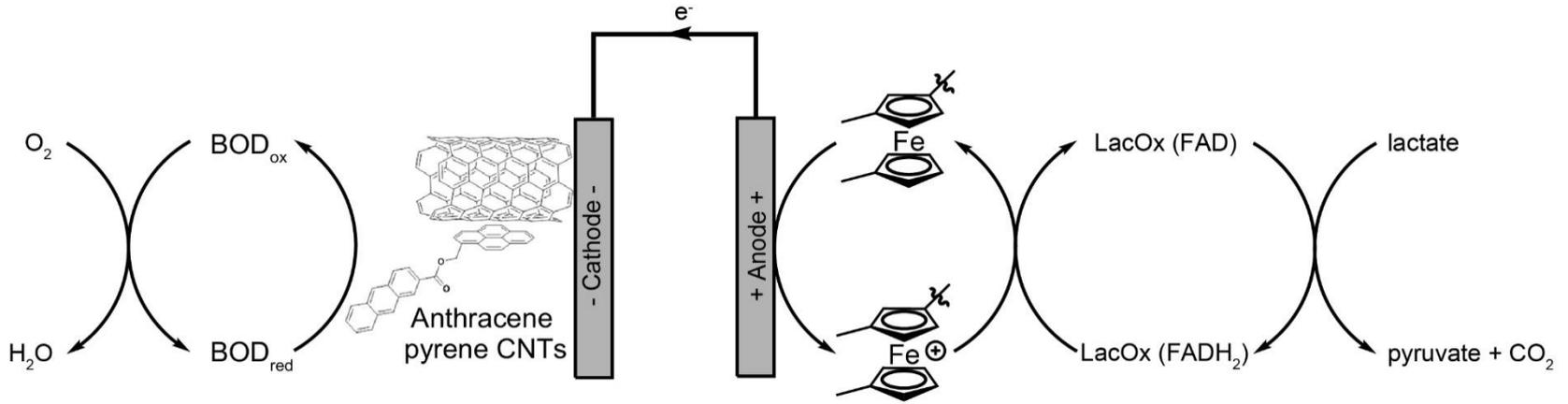
Surface Area



Conductivity

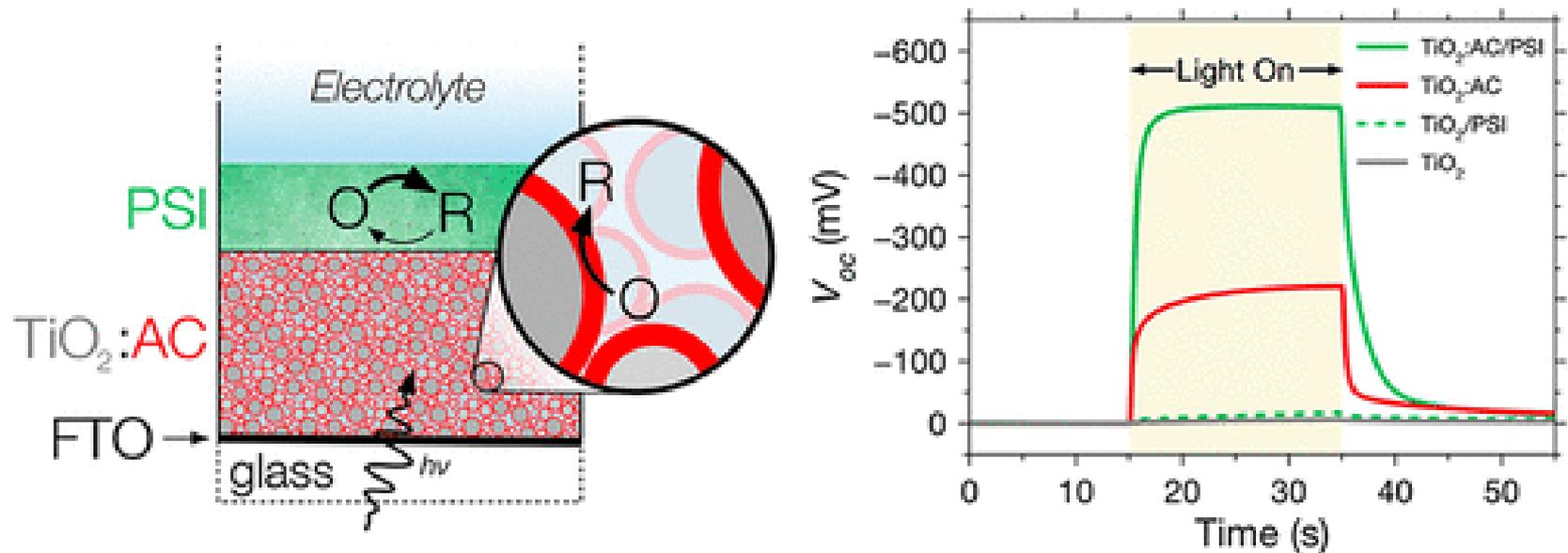


Prototype schematic and fabrication



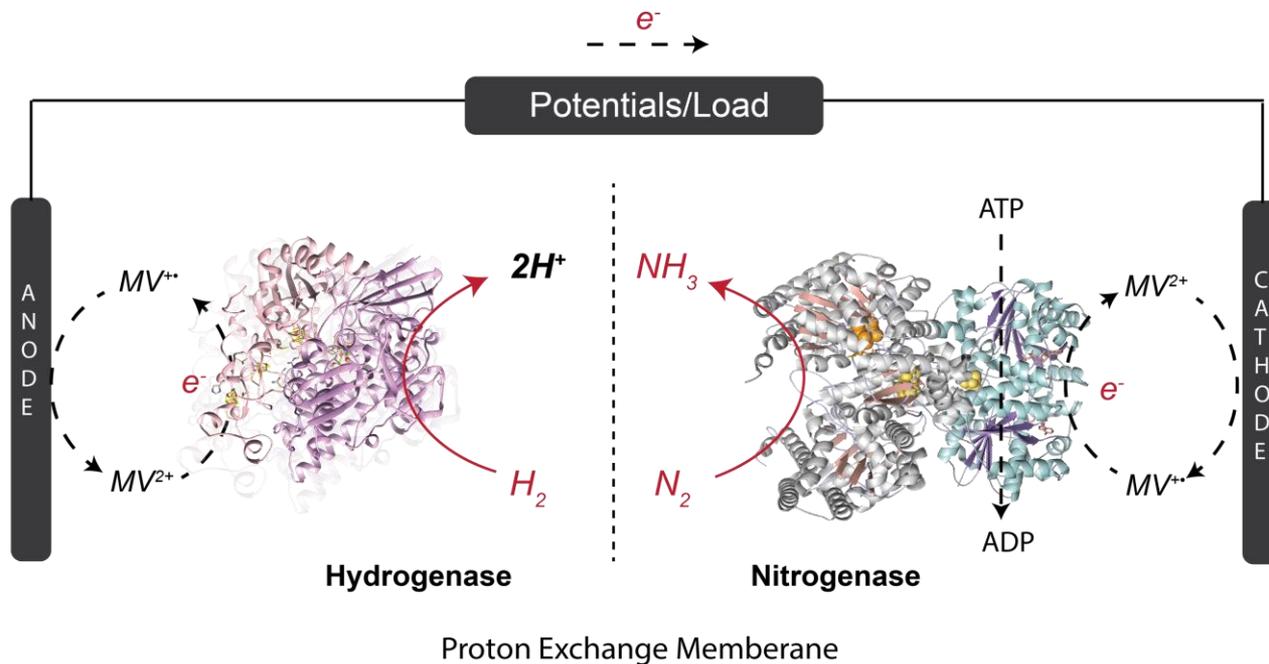
OCV: $0.443 \pm 0.08 V$
 J_{max} : $21.9 \pm 4 \mu A cm^{-2}$
 P_{max} : $2.37 \pm 0.9 \mu W cm^{-2}$ (0.5 μW)

Harvesting solar energy



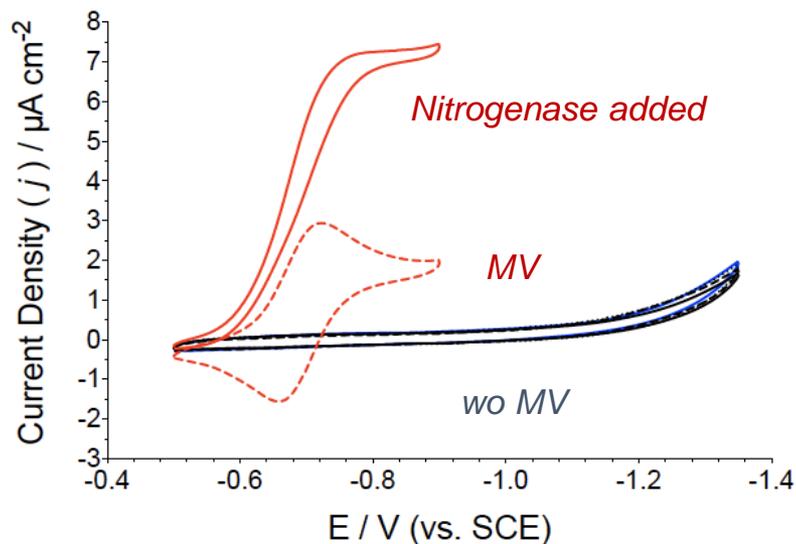
Dave Cliffl et al. *ACS Appl. Energy Mater.* 2018, 1, 2, 301-305

Bioelectrosynthesis



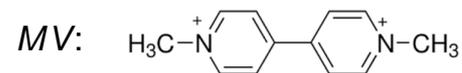
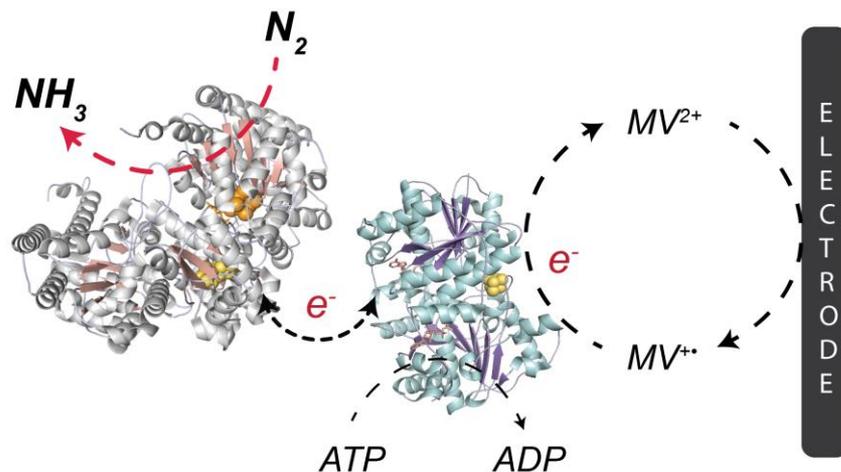
Nitrogenase Biocathode

Cyclic Voltammetry



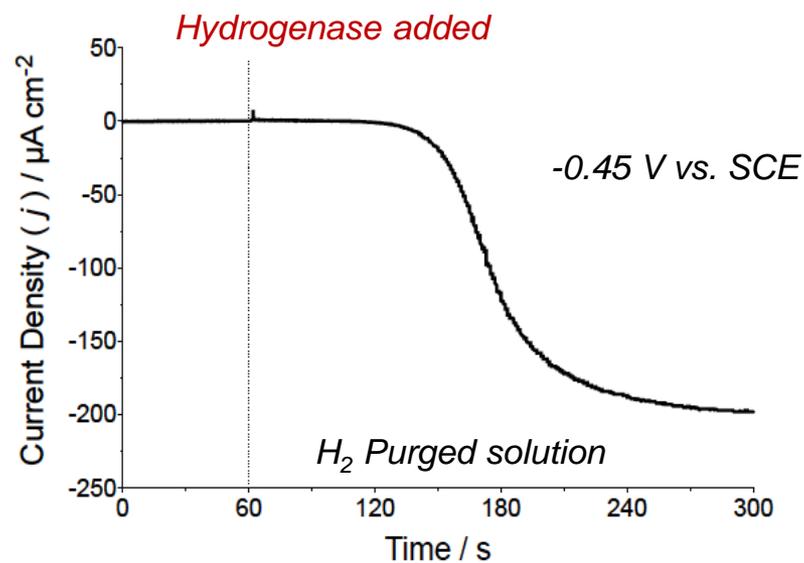
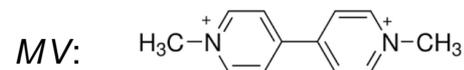
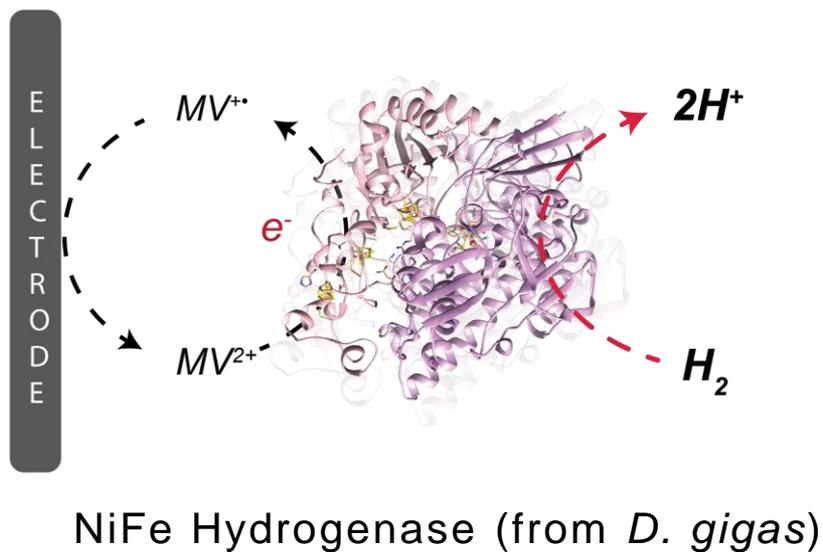
Bulk Electrolysis

Applied of -0.85 V vs. SCE for 1 hour



- $2.1 \pm 0.5 \mu\text{mol of NH}_3 \text{ mg}^{-1} \text{ MoFe}$
- Faradaic efficiency of $59 \pm 6 \%$.

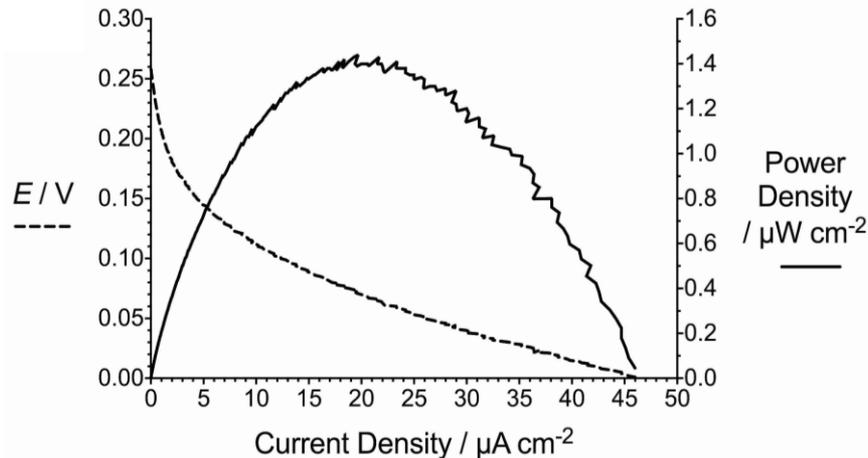
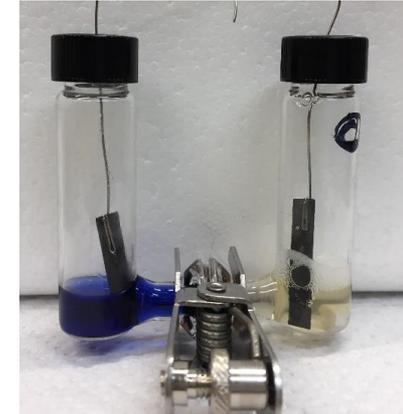
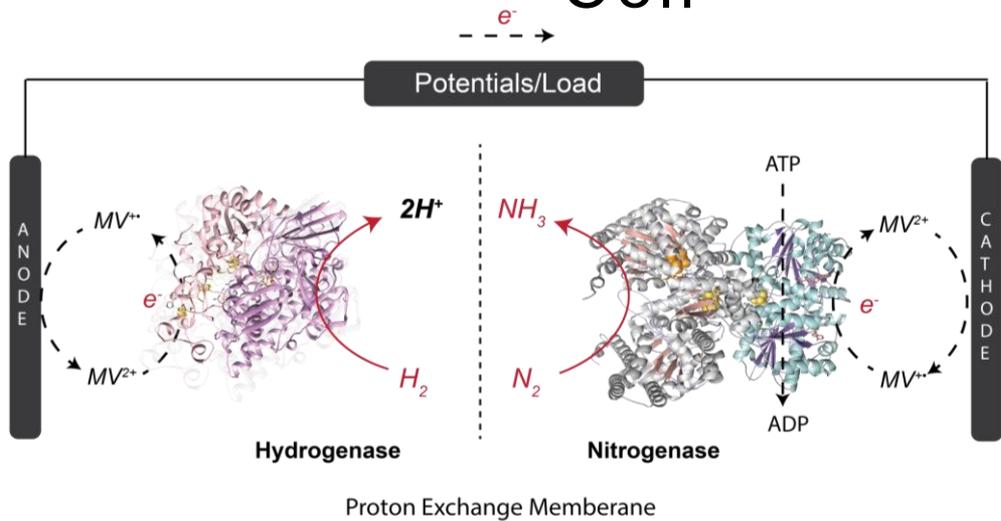
Hydrogenase Bioanode



Dr. Antonio De
Lacey

Amperometric $i-t$ analysis

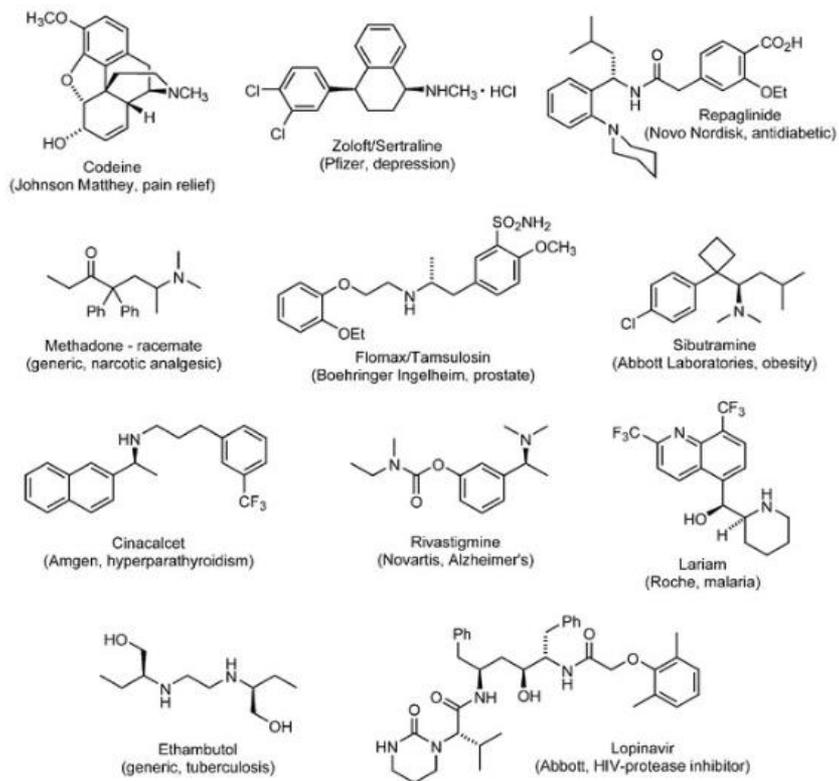
An Ammonia-Producing H₂/N₂ Fuel Cell



- OCP = 228 ± 28 mV
- $J_{\text{max}} = 48.0 \pm 3.1$ $\mu\text{A cm}^{-2}$.
- 286 ± 1 nmol NH₃ mg⁻¹ MoFe protein (60 mC)
- Faradaic efficiency of 26.4 ± 0.1 %.

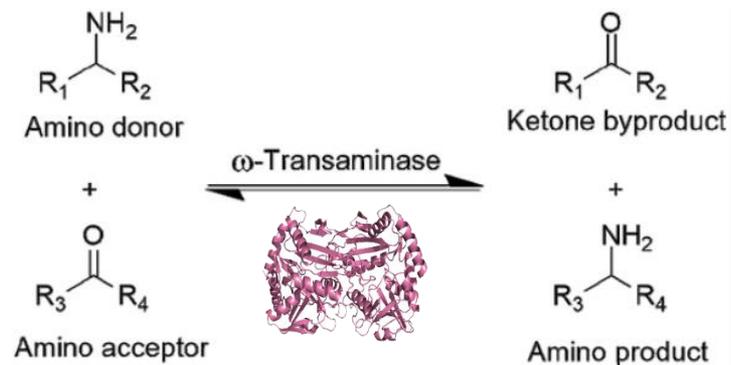
Chiral amines and ω -Transaminase (TA)

Examples of chiral amine drugs

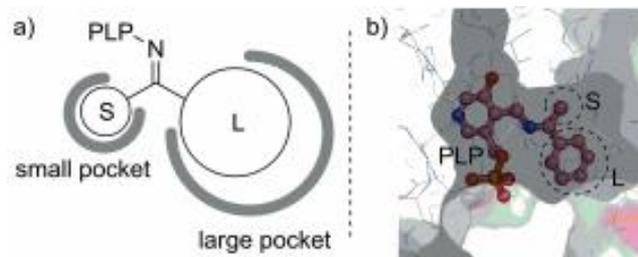


Nugent, T. C.; El-Shazly, M. *Adv. Synth. Catal.* 2010, 352, 753-819.

Reactions catalyzed by ω -TA



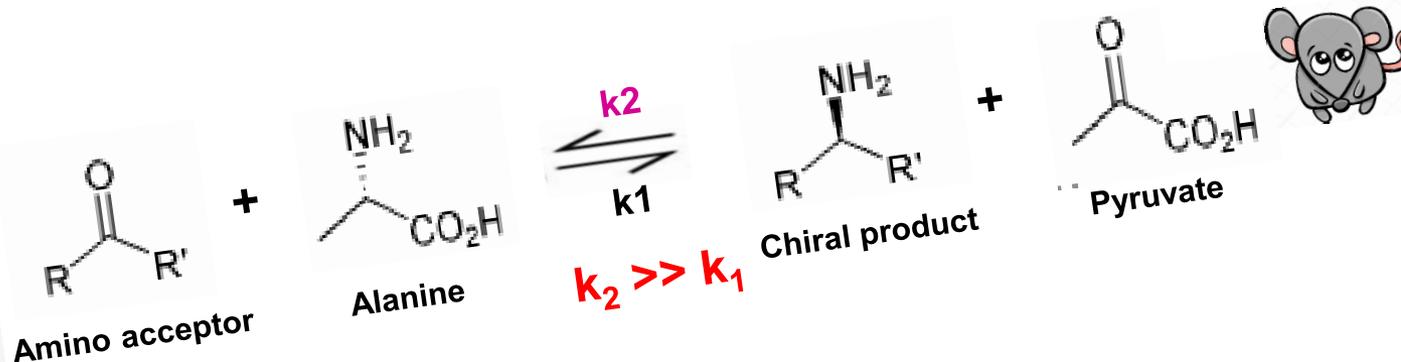
Chiral recognition mechanism



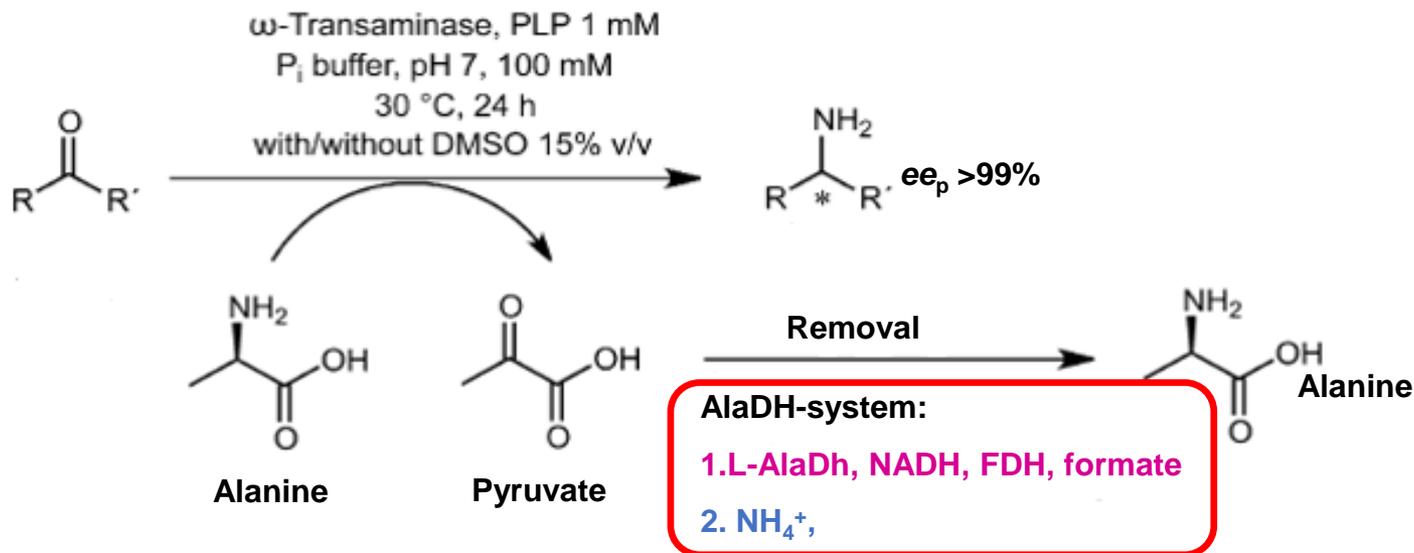
Fuchs, M.; Farnberger, J. E.; Kroutil, W. *Eur. J. Org. Chem.* 2015, 6965-6982.

Shifting reaction equilibrium

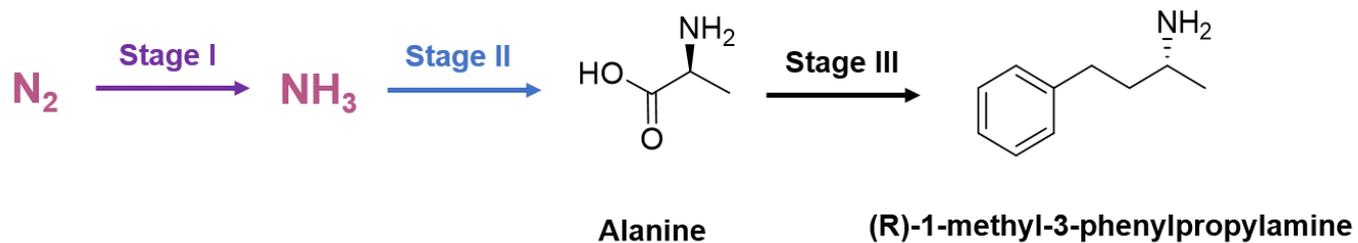
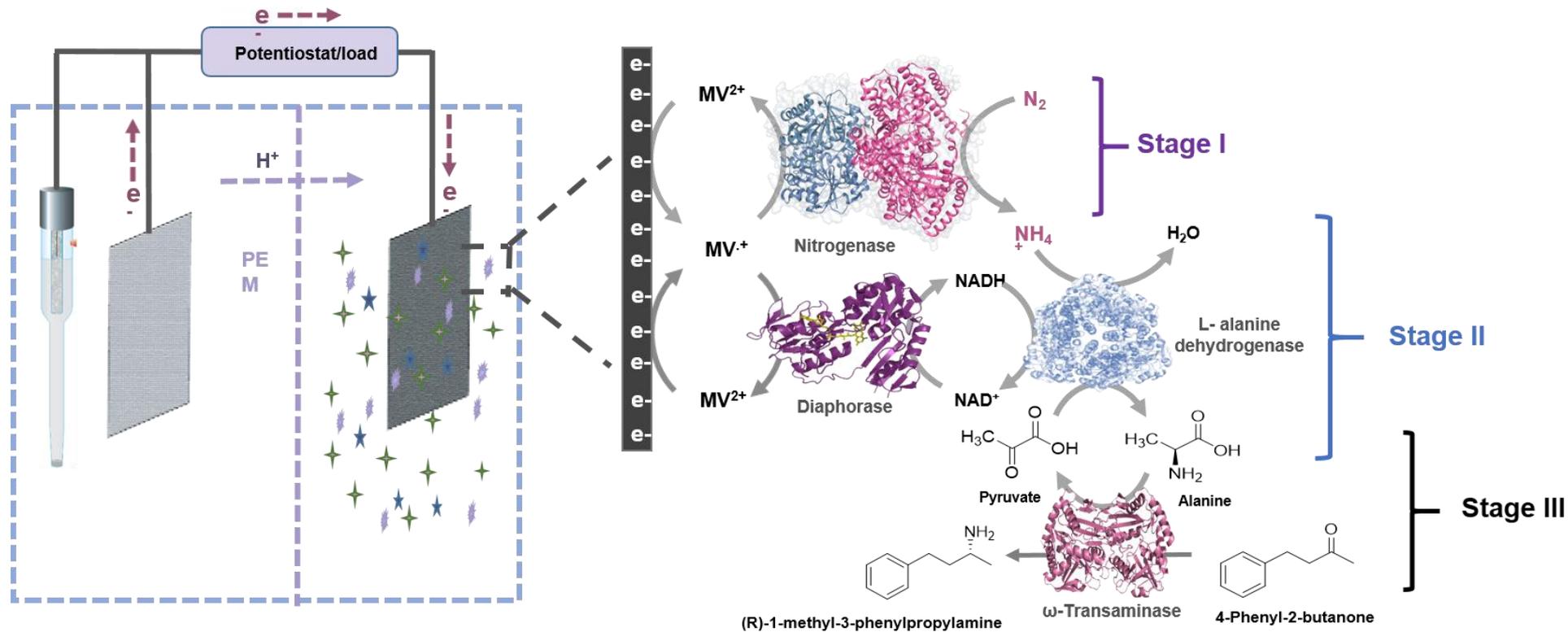
Equilibrium of reactions catalyzed by ω -TA



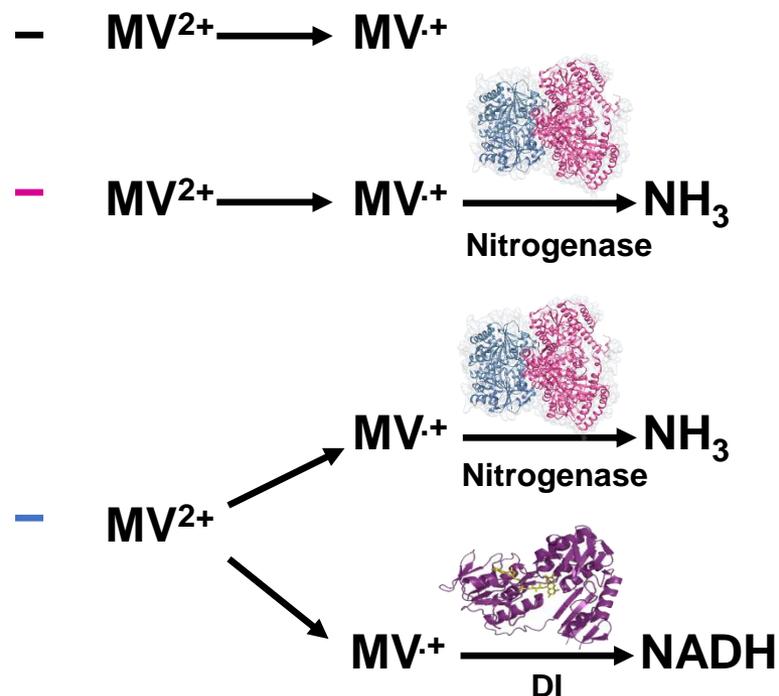
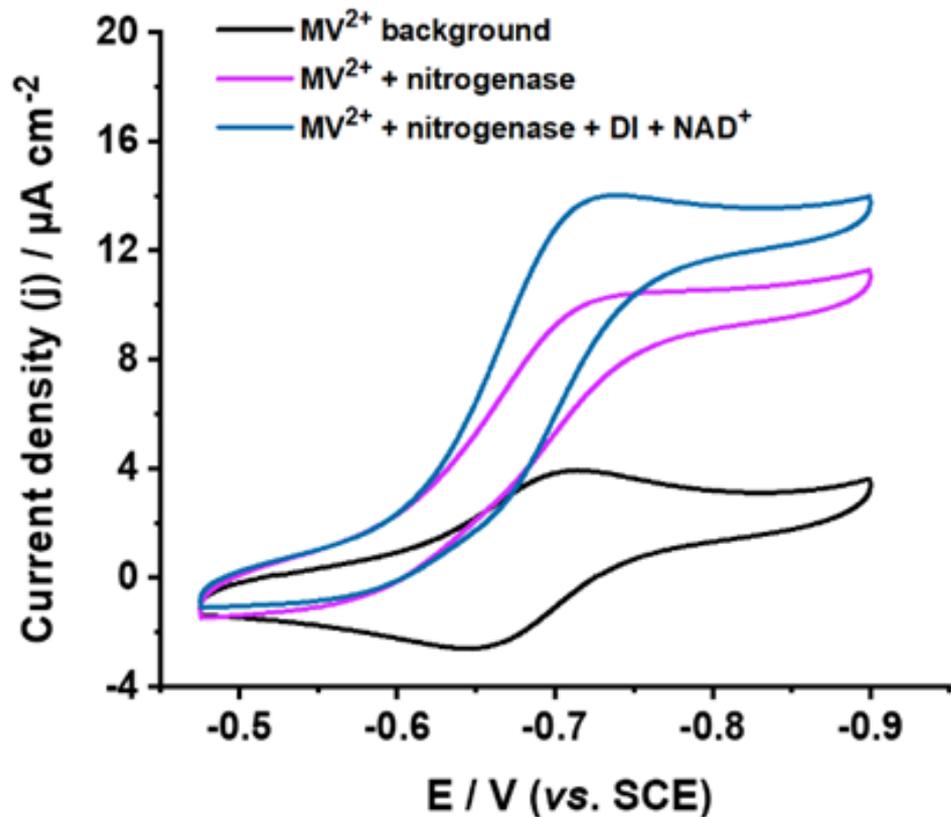
Alanine regeneration to push the equilibrium to the side of amine product



Upgraded N₂ fixation system based on bioelectrocatalysis

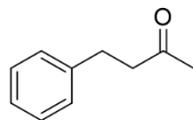


Bioelectrocatalysis using MV^{2+} as electron mediator

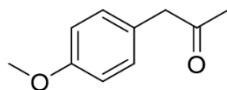


Applied potential for the bioelectrocatalytic system was set at -0.85 V vs SCE.

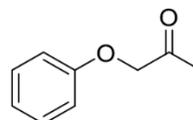
Bioelectrosynthetic reductive amination of substrates with different structures



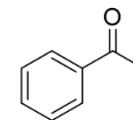
4-Phenyl-2-butanone (1a)



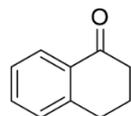
4-Methoxyphenylacetone (2a)



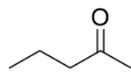
Phenoxyacetone (3a)



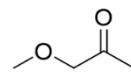
Acetophenone (4a)



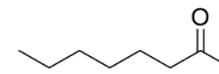
1-Tetralone (5a)



2-Pentanone (6a)



Methoxyacetone (7a)



2-Octanone (8a)

The prochiral ketones as the substrates of upgraded N₂ fixation system to produce chiral amines

Table: Asymmetric amination of various prochiral ketones catalyzed by upgraded N₂ fixation system

Entry	Substrate	Product concentration (mM)	ee _p (R) (%) ^[a]
1	1a	0.54	>99
2	2a	0.34	>99
3	3a	0.61	>99
4	4a	nd	nd
5	5a	nd	nd
6	6a	0.14	>99
7	7a	0.53	>99
8	8a	0.35	>99

Substrate concentration: 2mM; Reaction time : 10h

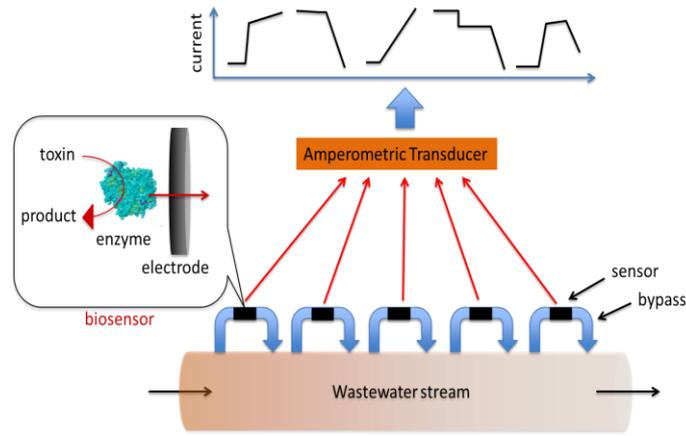
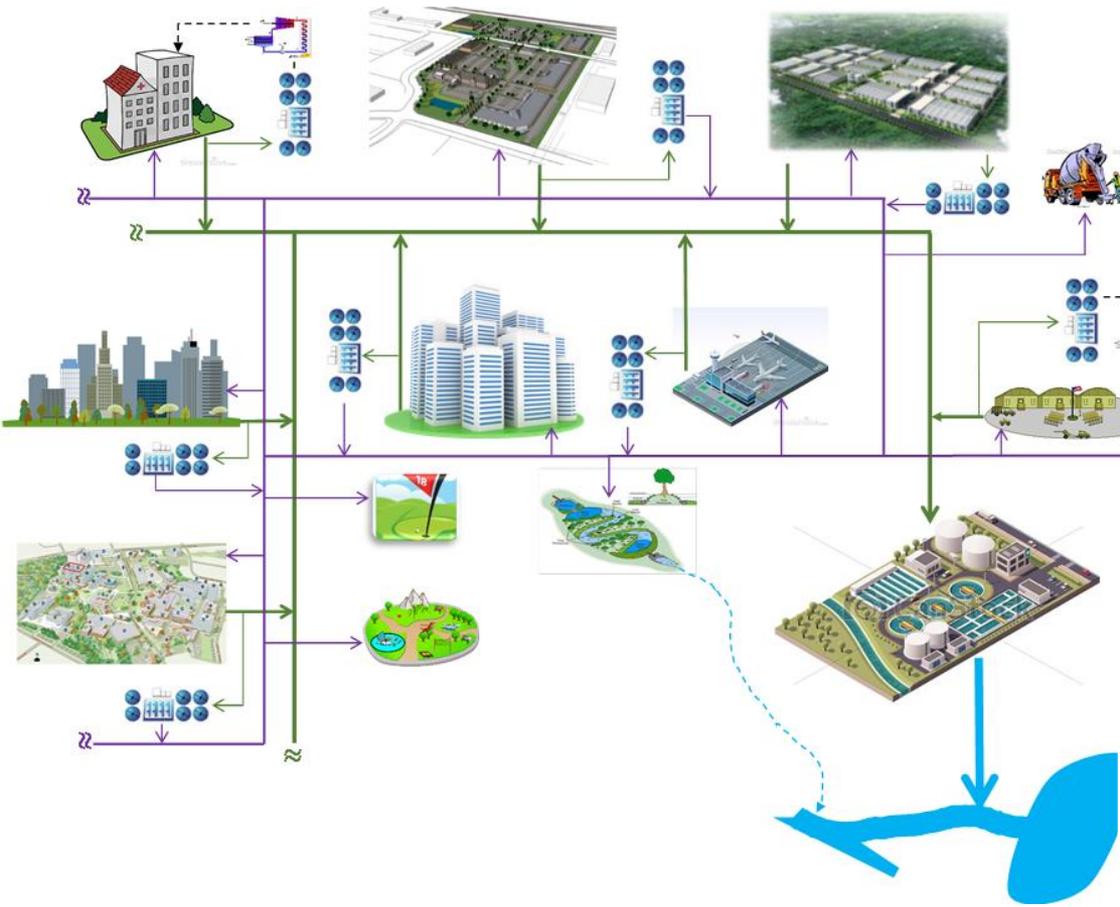
Future Challenges

- Understanding electron transfer pathways in microbes and enzymes – in-situ and operando techniques
- Materials to promote electron transfer pathways
- Synthetic biology and enzyme engineering
- Shelf life versus operational stability in a variety of environments
- Device engineering – bringing materials science/reactor design/synthetic biology together
- Integration into systems

Smart/Dynamic (Bio)sensors and (Bio)Electrochemical Systems

Recycle Water Networks

Visionary system of integrating bioreactors (including electrochemical ones) with sensors (including microbial biosensors) for motoring the “state of health” of the system and providing technological and environmental feedback control.



Conceptual Recycle Water Networks with interconnected networks of independent reuse water producers who buy and sell reuse water.

By Prof. Plamen Atanassov, University of California-Irvine and Michael Cooney, NHEI, University of Hawaii, Manoa, HI

- renewable water grid
- sewer grid
- biogas and on-site generated electricity

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